A Thesis for the Degree of Ph.D. in Engineering

Development of Portable Analytical Devices for Organophosphate Pesticide Detection

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Thesis Abstract

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Thesis Title:

Development of Portable Analytical Devices for Organophosphate Pesticide Detection

Thesis Summary

Substantial efforts have been made to develop enzymatic biosensors based on screen-printed electrodes and paper-based devices, which both are prominent approaches to qualitative and quantitative on-site analysis of pesticide residues. However, none of the hitherto developed biosensors has become available for practical applications due to limitations in enzyme stability, the requirement to perform multiple assay steps and the use of complicated and expensive methods for enzyme activity stabilization. To overcome these issues, the current work focuses on enzyme activity stabilization, the simplicity of sensor fabrication procedures and the reduction of the assay steps to be performed by the user.

Chapter 1 introduces the background and summarizes previous researches.

Chapter 2 describes the development of an amperometric enzymatic biosensor for organophosphate pesticide detection based on disposable screen-printed carbon electrodes (SPCE), surface-modified with gold nanoparticles (AuNPs), functionalized multi-walled carbon nanotubes (f-MWCNTs), chitosan and the acetylcholinesterase (AChE) enzyme. For electrochemical measurements, ferricyanide was used as a freely diffusible redox mediator in solution to eliminate interferences. AuNPs and f-MWCNTs contributed to biosensor sensitivity enhancement, while chitosan was applied for the stabilization of the AChE enzyme.

Chapter 3 describes the development of long-term stable flow control-based 3D microfluidic paper-based analytical devices (μPADs) for the single step analysis of organophosphate pesticides. Flow rate control was achieved by integrating a wax-patterned microfluidic channel into 3D paper-based devices. The slow movement of sample solution enables two simultaneous processes without requiring any user interaction: the inhibition reaction between the pesticide and the AChE enzyme, as well as the hydrolysis of indoxyl acetate (IDA) by the remaining active enzyme fraction, resulting in the colorimetric signal. AChE was physically adsorbed on skim milk-coated paper substrates to prevent non-specific reaction and to stabilize the enzyme. Finally, fully reagentless paper-based 3D devices with integrated pH buffering function were developed.

Chapter 4 describes the integration of electrospun membranes into the flow control-based 3D devices to further enhance the enzyme storage stability. The AChE enzyme was entrapped into an electrospun polyvinyl alcohol (PVA) fiber mat, which was then integrated into the 3D μPADs.

Chapter 5 summarizes the achieved results of this study.